In This Issue . . .

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The Puzzle of Cutaneous Touch

Being lucky in science depends upon the intersection of opportunity and preparation. It is through hard work that a scientist is "prepared" when the opportunity for a scientific breakthrough occurs. Dr. Gerald Krueger, Professor of Medicine (Dermatology) at the University of Utah, has diligently studied a variety of skin-grafting techniques in animal models, including several models that he has pioneered. The enormous work involved in these procedures has paid off in breakthroughs in skin pharmacology, immunology, and specific skin diseases. Now, Dr. Krueger, joining with Kathleen English, Naida Stayner, and Robert Tuckett of the Department of Physiology at the University of Utah, has applied one of his animal-model systems to the study of cutaneous innervation.

A major advance in the management of extensive burns has been the development of "skin equivalents," in which the subjects' own keratinocytes are greatly expanded in tissue culture, seeded onto an artificial dermis, and grafted onto the burn sites. Unfortunately, even with successful long-term grafts, touch sensations in such skin grafts are absent or severely reduced. In this study, cutaneous sensation was evaluated in rats who received autologous "skin equivalents," or skin equivalents with small pieces of normal skin, or "touch domes," included. Isolated peripheral nerves were attached to recording electrodes to detect afferent nerve impulses, and histologic nerve counts were performed on silver-stained frozen sections of grafted skin. The specialized areas within skin called "touch domes" are elevated dermal papillae covered by a thickened epidermis with Merkel cells in the basal epidermal layer. Touch domes were included in some grafts because they were felt to be a potential source of growth factors for regenerating type I sensory nerves.

The Utah investigators found that although skin equivalents became innervated by 83 days after grafting as demonstrated by silver staining, the functional sensory responses to light touch were poor. If either normal skin or a touch dome site was included within the skin equivalent, sensation was enhanced within these areas. Biopsy of these sites showed the presence of Merkel cells in the epidermis. If skin that did not originally contain touch domes was used for the skin equivalent graft, then no activation characteristic of type I afferent sensory nerve responses was observed. If the graft was of poor quality, sensory function did not return. It appears that reinnervation with type I sensory nerves is not enough for return of touch sensation and either the dermal papillae of the touch dome or the Merkel cell may be necessary for type I sensory activation.

Sites of Action of Retinoic Acid in Human Skin

Among the leaders in understanding the mechanisms of the profound effects of the retinoids on skin has been a group of dedicated investigators at the University of Michigan. They recently showed (JID 98:673–679, 1992) that retinoic acid differentially induces the cellular retinoic acid–binding receptor II (CRABP-II), but not CRABP-I, nor the nuclear retinoic acid (RAR-g) receptors. In this study, volunteers applied a single dose of retinoic acid under occlusion for 4 days, and then biopsies of treated and untreated skin sites were obtained.

The Michigan group applied a new molecular biology technique to localize the cellular site of retinoic acid action. They examined for the mRNA for each of the retinoid receptors using cRNA probes for hybridization to skin sections. Rather than labeling the probe with the traditional radioisotope, they used a nonradioactive signal, by end-labeling their probe with dioxygenin. Detection of the specific probe was obtained by an immunoperoxidase method, using a peroxidase-labeled anti-dioxygenin antibody, followed by a color reagent. This adaptation of the immunoperoxidase technique avoids the hazards of radioactivity and, in this study, produced excellent results.

The investigators observed that only the CRABP II receptor was induced by the application of retinoic acid to skin, and was localized equally throughout the epidermis, in endothelial cells and fibroblasts of the superficial dermis. Thus, the CRABP II receptor appears to be the key retinoid receptor induced in response to retinoic acid. Future studies by the Michigan group, headed by Dr. Voorhees, will focus on this receptor.

Is There Altered Elastogenesis in Buschke-Ollendorff Syndrome?

Of all inherited skin diseases, those characterized by defects in elastin are among the most rare. The combination of small skin thickenings at birth and asymptomatic spotting on bone radiographs (osteopoikilosis) is characteristic of the Buschke-Ollendorff syndrome, an autosomal dominant disorder of connective tissue. The skin lesions are considered to be connective tissue nevi and consist of accumulations of both elastin and collagen, with elastin deposits proportionately greater. In this issue, a multicenter collaboration between Vanderbilt University; the University of Texas, Houston; and the University of Washington resulted in a comprehensive analysis of connective tissue in two subjects with Buschke-Ollendorff syndrome.

A striking finding was that cultured skin fibroblasts obtained from the connective tissue nevi showed constitutive overproduction

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of elastin. Elastin mRNA levels were also elevated, suggesting a problem in regulation of elastin production. Increased sensitivity to transforming growth factor B-1 (TGFB-1), known to upregulate elastin, was not observed. The cytokine, basic fibroblast growth factor (bFGF), downregulated the production of elastin in the affected fibroblasts.

An obvious possibility is that these patients might have a mutation in the elastin gene, but no abnormalities could be found by restriction enzyme patterns in the subjects or their families. The exact reason for excessive elastogenesis in Buschke-Ollendorff syndrome remains unclear, but future studies focusing on regulation of the elastin gene should provide the needed breakthroughs. The investigative team included three members of the Department of Pathology at Vanderbilt (Drs. Gabriella Giro, Rita Kennedy, and Jeffrey Davidson); three from UT Houston (Drs. Madeleine Duvic, Ronald Rapini, and Frank Arnett); and Lynne Smith from the University of Washington, who provided the ultrastructural studies.

On the Origin of Mast Cell Enzymes

The human skin mast cell is situated just adjacent to superficial blood vessels, where it can modulate a number of inflammatory events. Mast cells contain a number of enzymes, including the four neutral proteases, tryptase, chymase, cathepsin G, and carboxypeptidase A. In this issue, investigators from the University of California San Francisco, directed by Dr. Sanford Goldstein, applied molecular biology techniques to examine one of the skin mast cell neutral proteases, carboxypeptidase A.

To determine whether skin mast cell carboxypeptidase A was unique to skin, they isolated a cDNA coding for the skin enzyme from dispersed dermal cells and compared its DNA sequence to that of lung mast cell carboxypeptidase A. They were identical. To examine the phylogeny of this enzyme, they compared it to bovine pancreatic carboxypeptidase A and found similar but not exactly the same substrate specificities for the purified proteins. Some of the substrate differences derived from the fact that human skin mast cell carboxypeptidase A, in contrast to bovine or rodent forms, is glycosylated. They developed an evolutionary tree and deduced that human skin mast cell carboxypeptidase A arose from gene duplication of carboxypeptidase B lineage rather than from pancreatic carboxypeptidase A. Understanding substrate specificity and the final structure of each mast cell enzyme will help clarify the function of this complex cell. Drs. Masaru Natsuaki, Caro-Beth Stewart, Peter Vanderslice, Megumi Natsuaki, William Rutter, and Bruce Wintrobe from San Francisco joined Dr. Lawrence Schwartz from the University of Virginia for this work.